

Correspondence

Phagocytosis and avirulent *Treponema pallidum*

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SIR—Uptake of avirulent *T. pallidum* (Nichols strain) was demonstrated in an *in vitro* system. Motile treponemes were harvested after culture for 7 days in spirolate broth (Izzat, Knox, and Wende, 1971). Non-motile treponemes were also obtained by keeping some of these at 4°C. for 18 hours. Macrophages were obtained from the peritoneal cavity of a rabbit by washing with 100 ml. Hanks's balanced salt solution (BSS) containing heparin (10 units/ml.). The collected macrophages were suspended in 2 ml. tissue culture medium 199 enriched with 15 per cent. rabbit serum (TCM-RS) containing treponemes (1 : 2, macrophages : treponemes). Identical aliquots of treponemes were added to 2 ml. TCM-RS without macrophages. These suspensions were incubated at 37°C. in an atmosphere of air with 5 per cent. CO₂. After 1 hour, the cell layer was re-suspended by gentle scraping, and dark-field counts were performed on the free treponemes. Electron microscopic examination was performed on macrophages following the methods of Sabatini, Bensch, and Barrnett (1963) and Palade (1952). Metabolic studies were performed on similar samples following the method of Musher, Keusch, and Weinstein (1972).

In five consecutive experiments, exposure to macrophages reduced the number of treponemes from an average of 15.3 to 8.6×10^6 per ml. Electron microscopic examination revealed macrophages containing segments of treponeme(s), consisting of a dense treponemal body surrounded by an envelope within phagocytic vacuoles. Some vacuoles contained treponemes with broken membranes. The metabolic study indicated that macrophages

incubated with motile or non-motile treponemes generated an increase in the amount of ¹⁴CO₂. This increase was statistically significant ($P < 0.001$), the motile treponemes-macrophages system showing an increase of 68.3 per cent. and the non-motile treponemes-macrophages system an increase of 40.9 per cent. This increase in percentage of decarboxylation of glucose by phagocytosing macrophages was obtained by comparison with resting macrophages and after subtracting the contribution by treponemes and background. Consequently, it was concluded that rabbit macrophages actively phagocytose avirulent *T. pallidum*.

Yours faithfully

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